

1                   **DIETARY FIBRE FROM BERRY-PROCESSING WASTE AND ITS**  
2                   **IMPACT ON BREAD STRUCTURE: A REVIEW**

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## Abstract

The structure and function of by-products of berry-processing industries are reviewed, with particular attention to dietary fibre (DF) and its effects in food products. The complex chemical composition and physicochemical characteristics of DF have been investigated and strategies for extraction of specific fractions that provide tailored technological and physiological functionality have been reviewed. The aim of this review is to describe in detail the **structural composition** and isolation methods of dietary fibre derived from berry by-products, and to explore their potential functionality in foods. The goal is to introduce DF from berry waste streams into the food chain, for which bread is a major vehicle. However, the appeal of bread lies in its aerated structure, for which DF is generally detrimental. The technological influence of DF on the formation and stabilisation of the aerated structure of bread is therefore reviewed, in order to understand how to incorporate DF into bread while maintaining palatability. The aerated structure of bread is stabilised by two mechanisms, the gluten matrix and the liquid film surrounding bubbles. Incorporating DF successfully into bread requires understanding its interactions with both of these mechanisms. DF fractions from berries offer superior nutritional value compared to cereal fibre, potentially with less damage to bread structure, due to the higher proportion of soluble fibre. By-products from berry-processing industries could be used as a source of technologically and nutritionally distinctive DF to fabricate foods with enhanced nutritional value.

*Keywords: dietary fibre, berries, by-products, fractionation, bread*

## 1. Introduction

The term “dietary fibre” (DF) has been evolving for more than fifty years. Early definitions described dietary fibre as the non-digestible constituents of plant cell walls (polysaccharides, lignin and associated substances).<sup>1</sup> Since then, the definition has been largely expanded and modified in response to new findings in nutrition and physiology as well as in advances in analytical techniques. The internationally recognised description of DF was introduced in 2009 (Codex Alimentarius 2009) with the aim to align the definition among countries and scientists.<sup>2</sup> Codex Alimentarius defines DF as carbohydrate polymers with ten or more monomeric units, which are not hydrolysed by the endogenous enzymes in the small intestine of humans, and which belong to the following categories: a) edible carbohydrate polymers naturally occurring in the food as consumed; b) carbohydrate polymers obtained from food raw materials by physical, enzymatic or chemical means and which have a beneficial physiological effect demonstrated by generally accepted scientific evidence; and c) edible synthetic or modified carbohydrate polymers that exhibit beneficial health effects that are demonstrated by generally accepted scientific evidence. The group of non-digestible carbohydrates includes cellulose,  $\beta$ -glucan, hemicelluloses, pectin, inulin, resistant starch and various oligosaccharides.

The Codex Alimentarius definition primarily focuses on composition of carbohydrate polymers and their associated health benefits, rather than the physicochemical properties of biopolymers. DF structures possess unique physicochemical characteristics (e.g., viscosity enhancement, water-holding capacity, bulking ability and fermentability) that are in turn accountable for differences in functional properties, technological applications and physiological responses.<sup>3</sup> DFs are subdivided into two general groups, soluble dietary fibre (SDF) and insoluble

97 dietary fibre (IDF) based on their water solubility.<sup>4</sup> It should be stressed that there is a  
98 clear distinction between “chemical solubility” and “nutritional solubility”. For  
99 example, arabinoxylans after aqueous extraction at high pH (~12) with continuous  
100 stirring for several hours at high temperatures (>60°C) are easily solubilised and are  
101 classified as “SDF”.<sup>5,6</sup> However, during digestion of unrefined berry fibre or cereal  
102 bran, the gastrointestinal tract (GI) does not provide the environment required to  
103 extract arabinoxylans efficiently, such that they remain in the IDF fraction of the  
104 fibre. Thus, arabinoxylans in their native state, as part of the plant structures, will not  
105 provide the functional or physiological responses associated with soluble fibres in  
106 contrast to the extracted arabinoxylan fractions.

107 SDFs form viscous solutions and are resistant to digestion in the small  
108 intestine, but easily fermented by the microbiota of the large intestine. The viscous  
109 and fermentable nature of SDF has been associated with a number of beneficial health  
110 effects, such as reduction of the glycaemic response and plasma cholesterol,  
111 prolonged gastric emptying, and slower transit time through the small intestine.<sup>7</sup> IDFs  
112 are generally porous and low-density structures, for which fermentation and solubility  
113 are limited in the human GI tract. IDFs include cellulose, some hemicelluloses,  
114 resistant starch and lignin. Health benefits associated with IDF are typically linked to  
115 laxative properties involving increase in faecal bulk and decrease in intestinal  
116 transit.<sup>8,9</sup>

117 Solubility could be used as an index to predict approximately the  
118 technological impact of a given dietary fibre on a food product. For instance, SDF  
119 may be desired in liquid formulations (e.g., soft drinks); however, at physiologically  
120 relevant levels, the resulting high viscosity is likely to detrimentally impact on the  
121 sensory characteristics of the product. By contrast, IDF can be formulated relatively

easily into solid or semi-solid foods (e.g., bakery or dairy), posing different technological challenges that need to be tackled, such as loaf volume reduction in leavened bakery products. However, classification of DF based exclusively on solubility may not be sufficient to differentiate between various functional properties and physiological responses, and a fuller classification may encompass particle size and molecular mass.<sup>10</sup>

Alternatively, DF can be classified according to sources, e.g. fruits, vegetables, cereals, legumes, nuts. The source of DF has a direct impact on the ratio of insoluble-to-soluble dietary fibre and hence its technological applications in foods. The ratio of insoluble-to-soluble dietary fibre differs between fruits and cereals, with lower levels of soluble fibre in the latter.<sup>11-16</sup> Since most of the hypocholesterolemic effects reported for DF are associated with the fermentable and viscous carbohydrates that comprise its soluble fraction, fruit DF exhibit superior nutritional value compared with those from cereals.<sup>17,18</sup>

Fruit by-products are primarily generated by the juice and wine industries. Recently, there has been an increased interest towards utilisation and processing of berry by-products, driven by sustainability considerations as well as health, the latter primarily due to the high amounts of associated phytochemicals and dietary fibre.<sup>19-21</sup> Phytochemicals (i.e., bioactive compounds) found in common berries include polyphenols and high concentrations of flavonoids including anthocyanins and ellagitannins.<sup>22</sup> Such compounds may create difficulties in development of certain products due to colour formation; however, intelligent product development and marketing strategies may use this characteristic as an advantage, to distinguish and differentiate products, rather than a limitation.

Most berries are utilised in the food industry for juice production, which is directly consumed or further used as a bioactive ingredient in food formulations. Berry pressing results in the production of a by-product, usually termed “press cake” or “pomace”, which is primarily composed of berry skins (exocarp), seeds and stems, and accounts for around 20-25 % of the entire berry mass.<sup>20</sup> Berry pomace contains up to 70 % of the polyphenols originally present in the berries,<sup>23,24</sup> as well as large amounts of cell wall polysaccharides (e.g., pectins, cellulose) that can be utilised as a source of dietary fibre, plus functional bioactives (e.g., antioxidants) and natural colour compounds.<sup>25,26</sup>

Botanically, a berry is a fleshy fruit without a stone, produced from the ovary of a single flower. Grapes (*Vitis vinifera*), currants (*Ribes spp.*), bilberries (*Vaccinium spp.*), blueberries (*Vaccinium* sect. *Cyanococcus*), cranberries (*Vaccinium* subg. *Oxycoccus*) and gooseberries (*Ribes uva-crispa*) are classified as ‘botanical’ berries, whereas strawberries and raspberries are excluded from the term. The current review defines berries from a consumer, not a botanical, perspective and therefore encompasses literature on dietary fibre from ‘non-botanical’ berry sources. Table 1 lists commonly consumed or processed berries, and their typical DF composition. Although the values are not strictly comparable because of the different measurement methods used, some general compositional features and a preliminary comparison between them can be established. In general IDF dominates, but it is notable that blackcurrants and grapes are particularly rich in soluble fibre.

The discussion so far has established that there are nutritional drivers to increase DF contents in foods, and that the by-products of berry juice processing offer physiologically and technologically promising sources of DF for use in food products. The aim of this review is to describe in detail the intricate structures and isolation

methods of dietary fibre derived from berry by-products, and to explore their potential functionality in foods. Bakery products, in particular bread, are a major vehicle for delivering fibre and its health benefits to consumers; however, bread and other bakery products are distinguished by aerated structures that are in general damaged by fibre, but may be enhanced by certain fibre fractions. Therefore the review discusses fibre interactions with the aerated structure of bread, as the basis for identifying strategies for adding berry fibre fractions to bakery products in ways that maximise the benefits and avoid the damaging effects of fibre.

**Table 1.** Dietary fibre composition of berries and berries by-products of industrial interest.

Source	SDF	IDF	TDF	Reference
<b>Blackcurrant pomace</b>	250-300 <sup>a</sup>	~470 <sup>a</sup>	720-770 <sup>a</sup>	11
<b>Blackcurrant</b>	78 <sup>a</sup>	560 <sup>a</sup>	638 <sup>a</sup>	18
<b>Bilberry press cake</b>	69 <sup>a</sup>	520 <sup>a</sup>	589 <sup>a</sup>	27
<b>Bilberry</b>	58 <sup>a</sup>	580 <sup>a</sup>	638 <sup>a</sup>	18
<b>Raspberry pomace</b>	0.3 <sup>b</sup>	38.1 <sup>b</sup>	38.4 <sup>b</sup>	28
<b>Raspberry</b>	73 <sup>a</sup>	580 <sup>a</sup>	653 <sup>a</sup>	15
<b>Blueberry pomace</b>	~1 <sup>b</sup>	~49 <sup>b</sup>	50 <sup>b</sup>	28
<b>Blueberry puree</b>	24-43 <sup>a</sup>	106-155 <sup>a</sup>	136-190 <sup>a</sup>	29
<b>Blueberry</b>	16 <sup>a</sup>	~460 <sup>a</sup>	476 <sup>a</sup>	30
<b>Cranberry pomace</b>	0.8 <sup>b</sup>	57.9 <sup>b</sup>	58.7 <sup>b</sup>	28
<b>Cranberry fibre</b>	52 <sup>a</sup>	530 <sup>a</sup>	582 <sup>a</sup>	31
<b>Grape pomace</b>	7-108 <sup>a</sup>	164-637 <sup>a</sup>	173-745 <sup>a</sup>	32
<b>Chokeberry</b>	54 <sup>a</sup>	666 <sup>a</sup>	720 <sup>a</sup>	33
<b>Cherry</b>	12.2 <sup>b</sup>	87.8 <sup>b</sup>	100 <sup>b</sup>	34

<sup>a</sup> values are expressed as g kg<sup>-1</sup> of dry material. <sup>b</sup> values are expressed as %.

Section 2 details the structural characteristics of fibre from berry sources; Section 3 considers how berry pomace can be fractionated into its fibre components; and Section 4 considers fibre effects in bread, based on a model of the two principal bubble stabilisation mechanisms that are responsible for the aerated structure of bread.

## 2. Structural characteristics of dietary fibre from berry sources

DFs from berries are predominantly carbohydrate polymers (e.g., pectins, hemicelluloses and cellulose) plus lignin. Carbohydrate polymers adopt a wide range of conformations based on the anomeric configuration of monosaccharides, chain length and branching of the side chains, monosaccharide composition, and combinations of linkages between them. Due to this structural variability, DF is one of the most heterogeneous and diverse group of biomolecules, providing not only a spectrum of technological and physiological functionalities, but also challenges in classifying DFs and ultimately incorporating them into food formulations.

DFs are located in the plant cell wall, i.e. the structure that surrounds some cells providing structural rigidity to the plant. Figure 1 illustrates the structures of the major dietary fibres and their location within the cell wall. The middle lamella is the layer that follows immediately after the cell wall that essentially affixes adjacent cells together. Cell walls contain the majority of the IDF, primarily cellulose, hemicelluloses and lignin. Other compounds include pectin, cutin and other waxes, as well as various structural glycoproteins (e.g., arabinogalactan), with the exact composition depending on the botanical source, the location of the cell and the stage of growth of the plant. The middle lamella is rich in pectin and some structural proteins and provides the major source of SDF. The ratio of insoluble-to-soluble polysaccharides and their polymeric composition vary depending on the phylogenetic class of the plant, tissue, origin, developmental stage and environmental conditions during growth. Generally, primary cell walls of dicots (e.g., fruits and vegetables) and non-commelinoid monocots (e.g., aroids, alismatids and lilioids) contain abundant pectic polysaccharides and structural proteins, whereas hemicelluloses are present in limited amounts.<sup>35</sup> In contrast, commelinoid monocots (e.g., *Arecales*, *Commelinales*



and *Poales* (cereal grasses)) contain a higher percentage of cellulose and hemicellulose, and only negligible amounts of pectin and proteins.<sup>36</sup>

Cellulose is a linear polysaccharide composed of  $\beta$ -(1 $\rightarrow$ 4)-linked glucose monomers with individual chains aggregating to form microfibrils *via* hydrogen bonding (Figure 1). Cellulose chains can be ordered, forming crystalline regions, or disordered thus forming amorphous regions. Celluloses derived from berries show different crystallinity indices (%) and morphological features that are typically attributed to the composition of the raw cellulose, their pre-treatment and extraction methods. For example, micro-fibrillar, micro- and nano-crystalline celluloses have been extracted from different grape varieties and berry parts with crystallinity indices ranging from 54.9 to 74.9 %.<sup>37-39</sup> The high molecular mass and crystalline nature of cellulose result in poor solubility that can be improved through physical or chemical modifications (e.g., microcrystalline cellulose, methylcellulose, carboxymethylcellulose). Cellulose is resistant to digestive enzymes in the small intestine of humans, but is partially metabolized by the microflora of the colon into short-chain fatty acids.

Lignin is the second most abundant compound present in biomass and the only one based on aromatic rings. As illustrated in Figure 1, lignin fills the spaces between the polysaccharides in the secondary cell-wall, contributing a substantial portion (~20%) of the cell-wall.<sup>36</sup> Lignin is a branched cross-polymer that is composed of repeating phenylpropane units, originating from three aromatic alcohol precursors (i.e., monolignols), *p*-coumaryl, coniferyl and sinapyl alcohols (Figure 1). The phenolic substructures that originate from these monolignols are called *p*-hydroxyphenyl (H for coumaryl alcohol), guaiacyl (G for coniferyl alcohol) and syringyl (S for sinapylalcohol) moieties.<sup>40</sup> The distribution of phenolic subunits (H, G

and S) in the lignin structure depends on the plant species, with the highest amounts of H-units reported in lignin originating from monocot rather than dicot plants.<sup>41</sup> Previous studies detected only G units in lignins derived from grapes in contrast to those isolated from grape stalks that contained H, G and S units with molar proportions of 3:71:26.<sup>42,43</sup> Molecular mass of lignin varies considerably between plant species and is highly dependent on the extraction method. Typically, lignin of high molecular mass ( $5\text{--}400 \times 10^3 \text{ g mol}^{-1}$ ) is obtained with the sulphite pulping method, in contrast to the smaller lignins isolated using Kraft ( $1\text{--}5 \times 10^3 \text{ g mol}^{-1}$ ) or organosolv ( $0.5\text{--}3 \times 10^3 \text{ g mol}^{-1}$ ) processes.<sup>44</sup> Lignins extracted from berry sources such as grapes had small molecular mass of  $2.6 \times 10^3 \text{ g mol}^{-1}$ .<sup>42</sup> Most lignin is hydrophobic, with the exception being that obtained by the sulphite pulping method, which is soluble in water in a wide pH range.<sup>37</sup>

Hemicelluloses cross-link cellulose microfibrils *via* hydrogen bonding, resulting in formation of the cellulose-hemicellulose skeletal network (Figure 1).<sup>45</sup> Hemicelluloses are a heterogeneous group of branched or linear polysaccharides with molecular mass ranging between 22 to  $770 \times 10^3 \text{ g mol}^{-1}$  for those sourced from cereals and  $1059\text{--}1167 \times 10^3 \text{ g mol}^{-1}$  for hemicelluloses derived from berries.<sup>11,46,47</sup> This group of dietary fibre includes, but is not limited to, arabinoxylans, xyloglucans, glucomannans, galactomannans and  $\beta$ -glucans. The composition of hemicelluloses varies depending on the class of the plant. Xyloglucans, mannans and xylans are found in abundance in the edible and non-edible parts of dicot plants (e.g., fruits and vegetables), whereas arabinoxylans and galactomannans are major matrix hemicelluloses in monocot plants (e.g., cereals).<sup>36,48</sup> Fractionation and extraction studies of common berries (e.g., blackcurrants, bilberries and grapes) revealed that isolated hemicellulosic fractions were composed of xyloglucans, xylans,

galactomannans, mannans and acetylated glucomannans.<sup>11,23,39,49,50</sup> The common structural feature of hemicellulosic polysaccharides is that most have a continuous  $\beta$ -(1 $\rightarrow$ 4)-linked backbone, with the exception of  $\beta$ -glucan which has both  $\beta$ -(1 $\rightarrow$ 4) and  $\beta$ -(1 $\rightarrow$ 3) linkages. Xyloglucans composed of  $\beta$ -(1 $\rightarrow$ 4)-linked glucose units that can be substituted with xylose *via*  $\alpha$ -(1 $\rightarrow$ 6) linkages. Some of the xylose residues can be further substituted with galactose and fucose.<sup>51</sup> The backbone of arabinoxylans consists of  $\beta$ -(1 $\rightarrow$ 4)-linked xylan units with arabinose side chains linked *via*  $\alpha$ -(1 $\rightarrow$ 2),  $\alpha$ -(1 $\rightarrow$ 3), or  $\alpha$ -(1 $\rightarrow$ 5) linkages. Side chains of arabinoxylans may also contain galactose, glucuronic and ferulic acids.

Soluble hemicelluloses are resistant to digestion in the small intestine but are easily fermented by the microbiota of the large intestine and have been credited with several beneficial physiological effects such as control of blood glucose and insulin levels.<sup>52</sup> These health benefits are attributed to the high viscosity of hemicellulose solutions in the lumen of the gastrointestinal tract, resulting in slower rate of glucose absorption.

Pectin is found in substantial amounts in the primary cell wall and middle lamella of dicot plants, where the entire cellulose-hemicellulose network is embedded in a matrix of pectic polysaccharides that form a hydrated and cross-linked three-dimensional network. Berry pectins are heteropolysaccharides of variable molecular mass (29-132  $\times 10^3$  g mol<sup>-1</sup>) and backbone mainly composed of galacturonic acid residues bonded *via*  $\alpha$ -(1 $\rightarrow$ 4) glycosidic linkages.<sup>11,53</sup> The structural classes of any pectic polysaccharide involve homogalacturonan (HG), which is a linear chain of  $\alpha$ -(1 $\rightarrow$ 4)-linked galacturonic acids, rhamnogalacturonan I (RG-I), which consists of a backbone of alternating  $\alpha$ -(1 $\rightarrow$ 4)-galacturonic acid and  $\alpha$ -(1 $\rightarrow$ 2)-linked rhamnose units that may be substituted with arabinan and galactan side-chains, and

rhamnogalacturonan II (RG-II) (Figure 1). The structure of RG-II is highly complex with twelve different sugars and over twenty different linkages. Other pectic polysaccharides are xylogalacturonans (XGA) and apiogalacturonans, primarily found in the cell walls of aquatic plants, cotton seeds, watermelons, peas, apples and soybeans.<sup>54</sup> Pectic polysaccharides isolated from berry sources (e.g., blackcurrant, cherry, bilberry) using various extraction conditions are predominantly composed of homogalacturonan-rich pectins with exception for pectins extracted from blackcurrants that could also contain RG-II regions.<sup>11,55,56</sup> The carboxyl groups of pectins can be methyl- or acetyl-esterified and the degree of esterification is directly related to the technological and physiological functionality of the biopolymer.<sup>7,57</sup> Berry pectins are predominantly methyl-esterified and the degree of methyl esterification (DME) varies between various berry sources and processing conditions with low DME reported for pectins extracted from grapes, raspberry and strawberry, whereas intermediate and high DME pectins were isolated from blackcurrants.<sup>58-60</sup>

Advanced nutrition and functionality requires tailoring the composition and solubility of the dietary fibres. There are multiple approaches that can be used to extract fibre from plant sources depending on the quality and purpose of the targeted fibre. The basic isolation strategies to extract IDF and SDF from berries are described in the next section.

### **3. Fractionation strategies of dietary fibre from berries and berry by-products**

The protocols for fractionation of dietary fibre vary from source to source and the sequence of extraction steps typically depends on the final compound of interest (e.g., pectin, hemicellulose, lignin or cellulose) and/or expected functionality of this compound. Table 2 summarises isolation strategies, compositions, and yields of DF

that have been reported for berries and berry by-products, while Figure 2 illustrates a fractionation strategy for blackcurrant pomace into its individual dietary fibre components. Technological applications of IDF are determined by the ratio of celluloses, hemicelluloses and lignin, whereas those of SDF (e.g., pectin and some hemicelluloses) are dictated by their chemical composition (molecular mass, galacturonic acid and neutral sugar contents, etc.), which varies with source and extraction method. The development of a tailored fractionation protocol typically involves modification of existing and tested isolation methods for both SDF and IDF. Basic fractionation strategies can be further adjusted in order to obtain more refined and tailored compounds. Generally, fractionation of dietary fibre aims to serve multiple purposes such as isolation and quantification of biopolymer compounds and removal of undesirable cell wall components.

Separation of berry biomass into its individual components starts with the pre-treatment stage (e.g., milling of material, pre-extraction with acidic sodium chlorite, organic solvent, ultrasonic irradiation, or steam explosion) and removal of starch, proteins, lipids, waxes, phenolics and pigments. The selection of pre-treatment method depends on the type of biomass (e.g., pomace, press cake, full berry), desired final products, desired yield, quality, and structural characteristics of isolated fractions.<sup>61-63</sup> Milling is a common pre-treatment in fractionation of biomass that particularly impacts the extractability of soluble dietary fibre. Optimum particle sizes for berry pomace have been reported to be ~250-350  $\mu\text{m}$ .<sup>64</sup> Following pre-treatment, the extraction of SDF can be performed using conventional solvent-, microwave-ultrasound- or enzyme-assisted isolation methods. Isolation of pectins is typically performed using acid extraction (e.g., nitric, hydrochloric or sulphuric acid) at pH values ~2.0 and high temperatures (60-100°C). These extraction conditions result in

isolation of linear (high proportion of HG) pectins with a high degree of esterification (DE) that are widely applied in high-sugar food formulations. Physicochemical properties of pectins can be tuned through modification of extraction parameters that involve time-temperature combinations, solvent-to-solute ratio, type of extraction buffer, number of isolation cycles, and volume of organic solvent used for pectin precipitation.<sup>65,66</sup> Hemicelluloses are typically separated from biomass using alkali extractions (e.g., NaOH or KOH) at various time-temperature (1-15 h, 20-90°C) conditions.<sup>38,39,49,67,68</sup> Generally, increase in alkali molar concentration results in higher extraction yields of hemicelluloses.<sup>49</sup>

Cellulose and lignin are separated simultaneously from berry biomass after extraction of SDF. Cellulose is isolated in the form of a solid residue that is rinsed step-wise with acetic acid, water, ethanol and acetone, while lignin is recovered from the soluble residue by precipitation with acidified water. A direct isolation of cellulose from biomass is typically performed using chlorine dioxide (ClO<sub>2</sub>) or acidified sodium chlorite (NaClO<sub>2</sub>), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) or a mixture of acetic and nitric acids (organosolv).<sup>69,70</sup> When lignin is a component of primary interest in biomass, the extraction strategy typically includes sulphite-pulping, Kraft or organosolv methods, which result in isolation of structurally different lignins.<sup>43,71,72</sup>

Many researchers have determined the amount of pectin, hemicellulose, cellulose and lignin in dietary fibre from various sources using standard methods (e.g., enzymatic-chemical, enzymatic and non-enzymatic gravimetric methods), while a few studies have attempted isolation and fractionation of biomass into the individual components.<sup>73-75</sup> A comprehensive fractionation protocol of blackcurrant pomace into pectins, hemicellulose, cellulose and lignin has been performed using isolation techniques that combine hot buffer extractions and step-wise separation of the insoluble

lignocellulosic residues.<sup>11</sup> A similar fractionation protocol was used for processing of grape skins, with the focus being the isolation of the cellulosic fraction.<sup>38</sup> Some studies focused exclusively on the fractionation of freeze-dried bilberries, blackcurrants and raspberries into their SDF and IDF fractions following acidic extractions.<sup>18</sup> Other studies focused on the recovery of SDF from press cakes or pomaces of blackcurrants, bilberries, blueberries, raspberries and cranberries and their fractionation into the hemicelluloses, acid-soluble, chelating agent-soluble and dilute alkali-soluble pectins using sequential extractions.<sup>23,28,76</sup> Water-soluble polysaccharides have been separated from strawberry, blackcurrant and chokeberry pomaces using enzyme-assisted extractions.<sup>77,78</sup> More refined structures (e.g., xyloglucans) have been isolated from concentrated alcohol soluble solids of blackcurrants and bilberries using anion exchange chromatography fractionation.<sup>79,80</sup>

**Table 2.** Isolation strategies for soluble and insoluble dietary fibres from various berry sources.

Source	Extraction conditions	Extraction results	References
<b>Soluble dietary fibre (SDF)</b>			
<b>Bilberry, blackcurrant, raspberry</b>	Hot water extraction (HWE) at pH 2.0, 80 °C, 5 – 7 L of water per kg <sup>-1</sup> of berries.	Yields ranged from 34-250 g of pectin per kg of dried berries or berry pomace. Linear (HG-I) pectins were isolated.	18, 11
<b>Cranberry pomace</b>	HWE at 75 °C for 30 min, 1:5 ratio of ground pomace to hot water, initial pH.	Extracts contain 14 g of solids per kg of solution. Carbohydrates comprise 887 g kg <sup>-1</sup> of pomace extract.	81
<b>Grape Pomace (Pinot Noir)</b>	HWE at 100 °C and solute to solvent ratio 1:12, particle size <249 µm.	Yield was 100 g of SDF per kg of pomace. SDF were primarily composed of pectic polysaccharides.	64
<b>Grape pomace (Cabernet Sauvignon)</b>	Ultrasound-assisted (UA) extraction with citric acid (various temperature, time and pH conditions	Yields ranged from 32-294 g of pectin per kg of pomace. Linear pectins with Mw in the	65

	were applied). Solid-to-liquid ratio 1:10.	range of 111-205 g mol <sup>-1</sup> . DE (%) ranged from 20.1-61.2%.	
<b>Berry press of red/black currant, raspberry and elderberry</b>	HWE (80 °C, 8 h, pH 6.2) and microwave-assisted extraction (MAE, extraction time of 30 min).	Higher yields of pectins were obtained with MAE (594-837 g kg <sup>-1</sup> of berry pomace) as opposed to the HWE (413-738 g kg <sup>-1</sup> of berry pomace). Pectins had high flow behaviour index.	66
<b>Grape pomace (Cabernet Sauvignon)</b>	UA alkali extraction with 0.4-2.3 M KOH at 20 °C for 2.6-3 h and solid-to-liquid ratio 1:48-60 (g mL <sup>-1</sup> ).	Extraction yield was 79 g of hemicellulose per kg of grape pomace. Optimum isolation conditions resulted in extraction of 36 g of xyloglucans, 11 g of mannans and 12 g of xylans per kg of grape pomace.	49
<b>Grape skins (Touriga Nacional)</b>	Extraction with ammonium citrate for 1 h followed by solution acidification to pH 2.5 and precipitation with ethanol.	Complex mixture of hemicelluloses was isolated with acetylated glucomannans being the most abundant hemicellulosic polysaccharide.	39
<b>Insoluble dietary fibre (IDF)</b>			
<b>Grape skins (Touriga Nacional)</b>	Extraction with mixture of HNO <sub>3</sub> and ethanol (1:4 v/v) for 1 h.	208 g of cellulose per kg of dry grape skins. Isolated cellulose was 66.1% crystalline.	39
<b>Grape skins (Chardonnay)</b>	Organic solubles, pectins, hemicelluloses and polyphenols were removed prior to the isolation of cellulose. Extraction was performed with H <sub>2</sub> O <sub>2</sub> at 45 °C for 8 h (pH 11.5) followed by NaClO <sub>2</sub> treatment at 70 °C for 5 h (pH 3.0 - 4.0).	Total yield was 164 g of cellulose per kg of dry grape skins. Isolated cellulose was 64.3% crystalline.	38
<b>Grape pomace</b>	a) Extraction with mixture of formic acid/acetic acid/water (30/50/20 v/v), liquid-to-dry matter ratio (25:1) at	Total yield ranged from 400-410 g of lignin per kg of dry grape pomace. Guaiacyl units were detected in isolated	43



107 °C for 3 h. lignins.  
b) MAE followed by  
extraction with mixture of  
formic acid/acetic  
acid/water.

#### 4. Functional properties of dietary fibre isolated from berries in bread

The incorporation of dietary fibre into bakery products, particularly breads, has received much attention, increasingly so in recent years due to their potential use in formulation of functional breads such as gluten-free or fibre-fortified breads. A number of studies have focused on the incorporation of dietary fibre into cereal products, however, only limited information is available in the literature on the fortification of bread with dietary fibre from berries and berry by-products (Table 3).<sup>26,82,83</sup> It has been widely shown that fibre-rich by-products from the food industry may be incorporated into the food systems having a range of functionalities (e.g., partial substitution of flour, viscosity enhancers or gel forming agents). As noted above, functional properties of dietary fibre depend on plant source, isolation method, degree of processing, IDF/SDF ratio and particle size.<sup>84</sup> From a technological point of view, for example, IDF/SDF ratio influences dough rheological behaviour, development time and water absorption rate of the flour mixture.<sup>85,86</sup> From a nutritional point of view, the source of DF that is utilized for fortification of foods should have a ratio of IDF/SDF between 1:1 and 2:1 in order to yield the maximum health benefits.<sup>87,88</sup> In the case of berry pomaces, the proportion of the insoluble fraction is frequently greater than soluble.<sup>34 11</sup>

**Table 3.** The impact of berry and berry by-products on quality of dough and bread.

Source	% added	Matrix	Result	References
Blackcurrant	10,	Bread	Optimum concentration	at <sup>83</sup>

<b>pomace (particle size &lt;1mm)</b>	20, 30	dough	10%.	
<b>Red grape (<i>Pinot Noir</i>) and white grape (<i>Pinot Grigio</i>) pomace</b>	5, 10, 15	White bread	Breads had dark colour and reduced loaf volume at concentrations >5% w/w. Red GP (15% w/w) – fortified bread was significantly firmer than 5% w/w, control and white GP (5%, 10%)-fortified breads. Optimum concentration at 5% w/w.	89
<b>Red grape by-product</b>	4, 6, 8, 10	Sour-dough mixed rye bread	Increase in bread hardness at concentrations >6% w/w. No change in chewiness and springiness at 4% and 8% of GP, respectively. Optimum concentration at 6% w/w.	90
<b>White grape (<i>Emir</i>) pomace</b>	2, 5, 10	White bread	Negligible decrease in loaf volume at concentrations >2% w/w. Optimum concentration at 5% w/w.	91

399

400       The addition of fibre causes alterations in bread quality, mostly perceived as  
401 detrimental, including reduced loaf volume, sticky dough, firm and dark-coloured  
402 crumbs, and taste alterations.<sup>86,92,93</sup> The extent of changes depends on the fibre source  
403 and quality, and fibre-substituted breads can be improved by tuning the isolation  
404 conditions and physiochemical properties of the fibre. For instance, the treatment of  
405 IDF fraction with H<sub>2</sub>O<sub>2</sub> (which facilitates lignin removal) results in whiter IDF with  
406 improved softening and swelling characteristics; white breads formulated with  
407 alkaline H<sub>2</sub>O<sub>2</sub>-treated IDF at 10 % concentration had comparable loaf volume to  
408 control bread.<sup>94</sup> Alternatively, enzymatic treatment of fibre with hemicellulases and  
409 pentosanases can give structural modification of fibres that leads to improvement of  
410 dough and bread quality (e.g., larger loaf volume, softer crumb and retarded staling).<sup>92</sup>

411           The appeal of bread lies in its aerated structure, made possible by the unique  
412   gluten proteins of wheat that form a viscoelastic dough able to retain the fermentation  
413   gases produced by yeast.<sup>95</sup> Generally fibre, irrespective of the source, is detrimental  
414   to the creation of this aerated structure.<sup>96</sup> However, a tantalising and persistent theme  
415   of the literature is that some fibre fractions appear to offer the potential for beneficial  
416   effects on bread structure and quality. As early as 1944, Shetlar and Lyman reported  
417   that “bran must contain an ameliorating factor which increases loaf volume as well as  
418   a destructive factor which influences baking”, and found that water extracts of bran  
419   increased loaf volume, while washed bran gave lower loaf volumes than the original  
420   unwashed bran.<sup>97</sup> By contrast, more recent research demonstrated the opposite effect,  
421   that aqueous extracts of bran decreased loaf volume, while the residual bran increased  
422   loaf volume compared with the white flour control, with the coarse bran residue  
423   giving the greatest benefit.<sup>98</sup> The effect of particle size similarly throws up contrasting  
424   results in different studies, with grinding bran to give small particles sometimes a  
425   benefit, sometimes more damaging.<sup>99</sup> When considering potential bread ingredients  
426   from other fibre sources such as berry pomace, resolving the puzzle of which fractions  
427   may be beneficial, and maximising the benefits, and which fractions may be  
428   detrimental, and then minimising or eliminating the damage, remains a key challenge.

429           Most studies on the effect of fibre on baked goods have focussed on cereal  
430   brans, and these studies are instructive for predicting or interpreting effects of other  
431   fibre sources such as berries, particularly in relation to understanding mechanisms of  
432   action.<sup>82</sup> Generally, bran damages the aerated structure of bread by means of three  
433   major mechanisms: gluten dilution, physical disruption of gluten films, and  
434   competition for water. It should be noted that fruit fibres show similar effects, but

their higher soluble fibre content compared with cereal brans alters the details of the effects.

An important paradigm for understanding effects of fibre on bubble stability and the creation of aerated structure in bread is what MacRitchie calls “the dual mechanism for bubble stability”.<sup>100</sup> It is well understood that the viscoelastic gluten matrix capable of retaining gases is at the heart of the unique appeal of bread and the unique importance of wheat.<sup>87, 92</sup> However, in addition to gluten-mediated retention of gases, there exists around bubbles in an expanding dough a thin liquid film that imparts a secondary stabilisation mechanism at the later stages of expansion.<sup>92,101,102</sup> As bubbles begin to come into contact during the later stages of proving and early stages of baking, discontinuities in the gluten-starch matrix arise that would normally cause exchange of gas leading to coalescence; however, the presence of the liquid film fills these discontinuities to provide some additional stability and allowing for longer expansion, a larger loaf volume, and a finer cell structure. This additional stabilisation mechanism helps to explain differences in bread making quality that are not accounted for by gluten quality, and provides a basis for understanding effects of surface active proteins and lipids and, for our current purposes, some of the effects of fibre addition.

Figure 3 illustrates and summarises these two mechanisms of bubble stabilisation in bread making and the factors that affect them, and provides the starting point for understanding effects of fibre. Firstly, the principal mechanism of bubble stabilisation in bread dough is the gluten-starch matrix. Broadly, addition of fibre disrupts the gluten, not only through dilution of the gluten and through competition for the available water, which can be addressed through addition of extra gluten and water, but also through direct interactions between fibre and gluten protein. A recent

review <sup>99</sup> confirmed the hypothesis <sup>103-106</sup> that fibre negatively affects the formation and physical properties of the gluten network through a combination of physical and chemical mechanisms, both of which are affected by particle size. In conclusion, both soluble arabinoxylans and insoluble solids from wheat bran have the same negative effect on gluten formation, mediated through viscosity and physical competition for water, and through chemical interactions *via* ferulic acid which allows crosslinking between arabinoxylans and gluten proteins, thus changing the agglomeration behaviour and formation of gluten network. Reducing particle size increased the negative effects by increasing the surface area for direct ferulic acid interactions and for diffusion of components (including ferulic acid) out of the bran particles. It has been also noted the greater water binding capacity of the insoluble fraction of bran and the benefit of xylanase in ameliorating the competition for water from this fraction.<sup>104</sup> Pectin effects in model bread systems similarly revealed a ferulic acid-mediated interaction between pectin and gluten proteins, weakening the formation of the gluten network.<sup>107</sup>

Less attention has been paid to the liquid film and the effects of fibre on this stabilisation mechanism. Additional gas cell stabilisation *via* the liquid film is particularly important in non-wheat doughs, which lack the primary gluten stabilisation mechanism.<sup>108</sup> The liquid film is stabilised directly by surface-active proteins and lipids, and indirectly by non-starch polysaccharides (NSP), the latter by increasing viscosity and thus slowing draining and retarding coalescence.<sup>109</sup> NSP can also exhibit interfacial activity due to the presence of functional groups (methyl and acetyl groups), ferulic acids or branched nature of their side chains. Alternatively, they can directly interact with proteins adsorbed at the interface, potentially modifying their surface-active properties.<sup>108</sup> Meanwhile a somewhat overlooked

component is lignin, which is a component of fibre that frequently accompanies NSP extracts and that has its own surface-active properties that can stabilise bubbles.

The literature on the effects of fibre has resulted in different hypotheses to explain the effects of bran on bread quality, with interpretation complicated by variation in definitions of bran and in its composition and physical properties as well as variations in bread making methods and in compensations made (such as the extra water added).<sup>99</sup> Understanding and hence optimising addition of fruit-derived fibres similarly requires an awareness of this complexity of effects. Crucially, most of the mechanistic studies to understand fibre effects have focussed on wheat bran, such that there is much to be done to elucidate with precision the effects of non-cereal fibres, particularly those lacking ferulic acid which appears to dominate wheat bran effects. The dual mechanism paradigm of bubble stabilisation illustrates that there is scope to identify soluble fibre fractions from fruit that enhance the viscosity-mediated benefit of liquid film stabilisation while avoiding the (largely) ferulic acid-mediated damage to the gluten matrix.

## **5. Conclusions**

Berry DF presents technological and physiological advantages compared to other sources of DF due to the lower ratio of insoluble-to-soluble DF, thus offering superior nutritional value with distinctive technological properties. To extract fibres with optimal functionality requires creating extraction protocols that target the dietary fibre fraction of interest to obtain refined and tailored compounds. Bread and bakery products are key vehicles for delivering fibre to the population, but dietary fibres are generally detrimental to the creation of aerated bread structure, although there is scope to identify fibre fractions that enhance bread quality. The effects of fibre on bread structure are mediated through the dual mechanisms of bubble stabilisation:

gluten-fibre interactions with the gluten network, and interactions of fibres with the liquid film that surrounds and stabilises bubbles during proving and baking. The drive to add value to berry processing wastes by using them as nutritionally beneficial and functionally promising food ingredients is pressing, with several research and practical issues to be resolved in order to identify, target and exploit berry fibre fractions in food products successfully.

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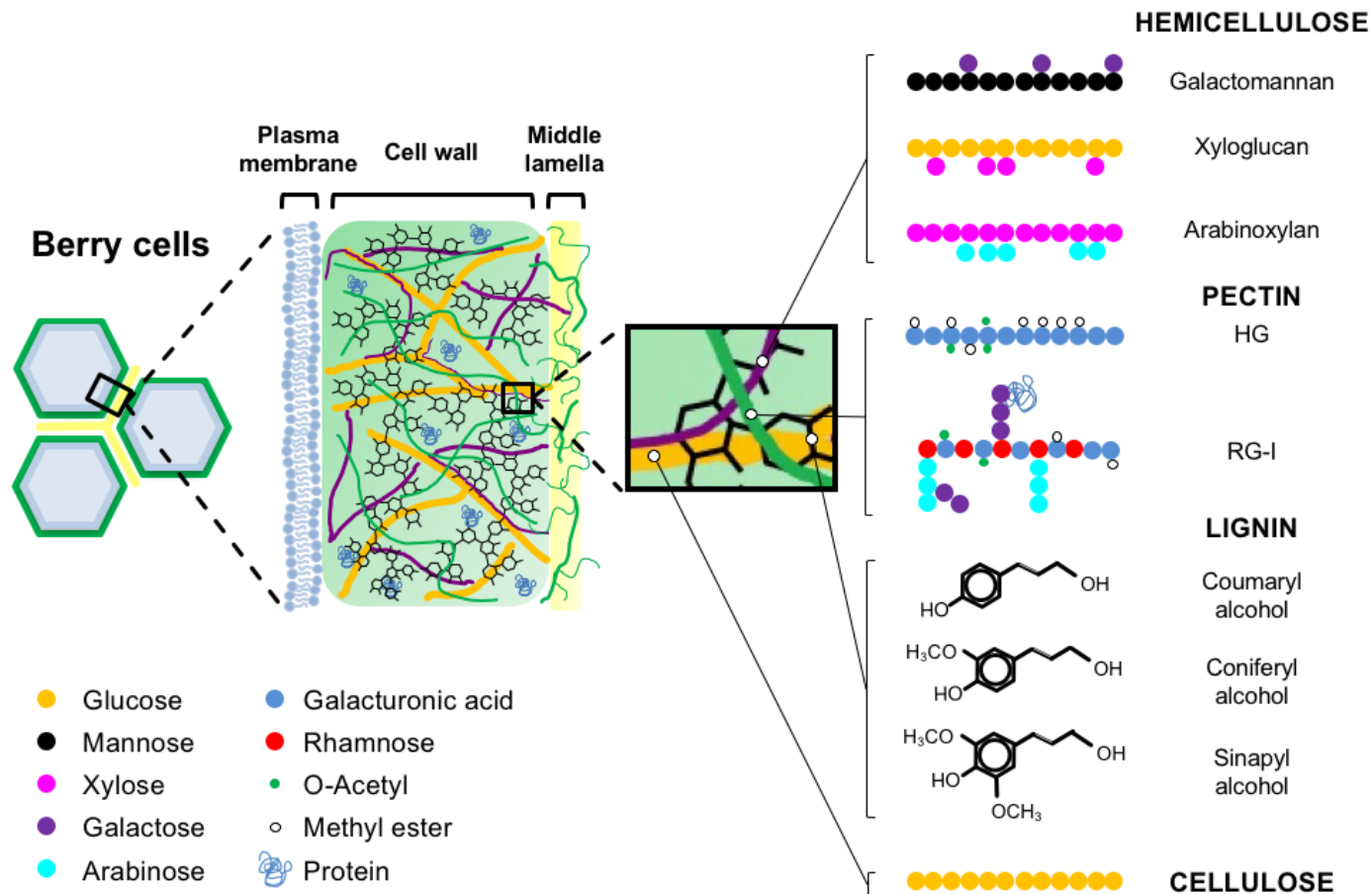
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## FIGURE LEGENDS

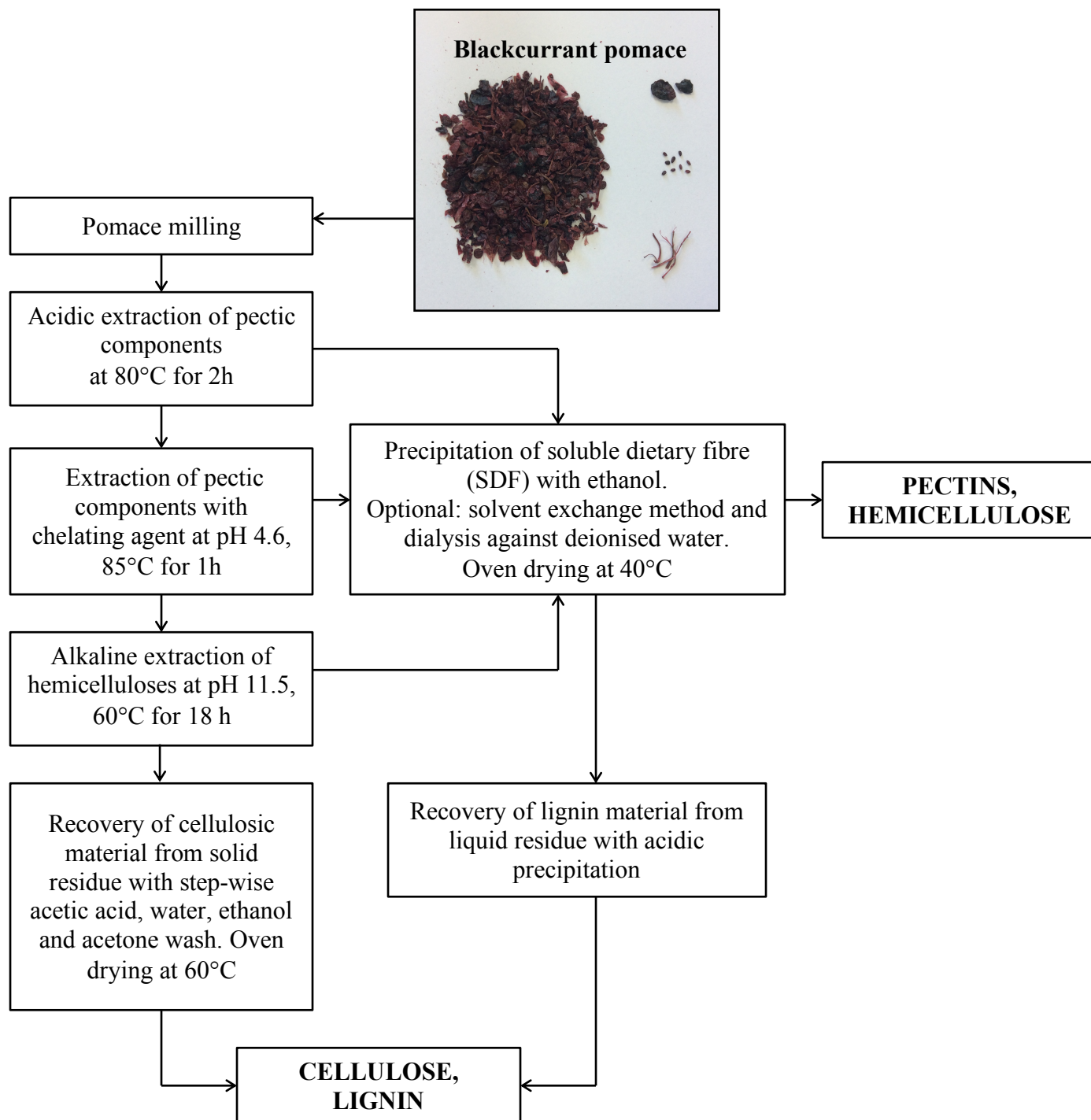
**Figure 1:** Location of dietary fibres within the cell wall and structures of the major dietary fibres. Dietary fibres are located in the cell-wall and in the middle lamella. The cell wall consists predominantly of insoluble- whereas middle lamella of soluble- dietary fibre. The sugar residues or alcohols in the case of lignin are also shown for each major structure.

**Figure 2.** Example of a basic extraction strategy used for fractionation of blackcurrant pomace into the individual dietary fibre components – pectins, hemicellulose, cellulose, and lignin. Adapted with modifications from Alba et al. (2018).

**Figure 3:** Illustration of the effects of fibre on the dual mechanisms for bubble stabilisation in expanded bread doughs. Top: Gluten-fibre interaction with the gluten network. Physical and chemical interactions between gluten and fibre generally have negative impact on bubble stability, worsened by reducing particle size. Bottom: Interactions with the liquid film. There is an indirect, positive effect of soluble fibre on stability of the film *via* increased viscosity, with direct physical interaction from insoluble fibre particles that may positively or negatively influence film stability.

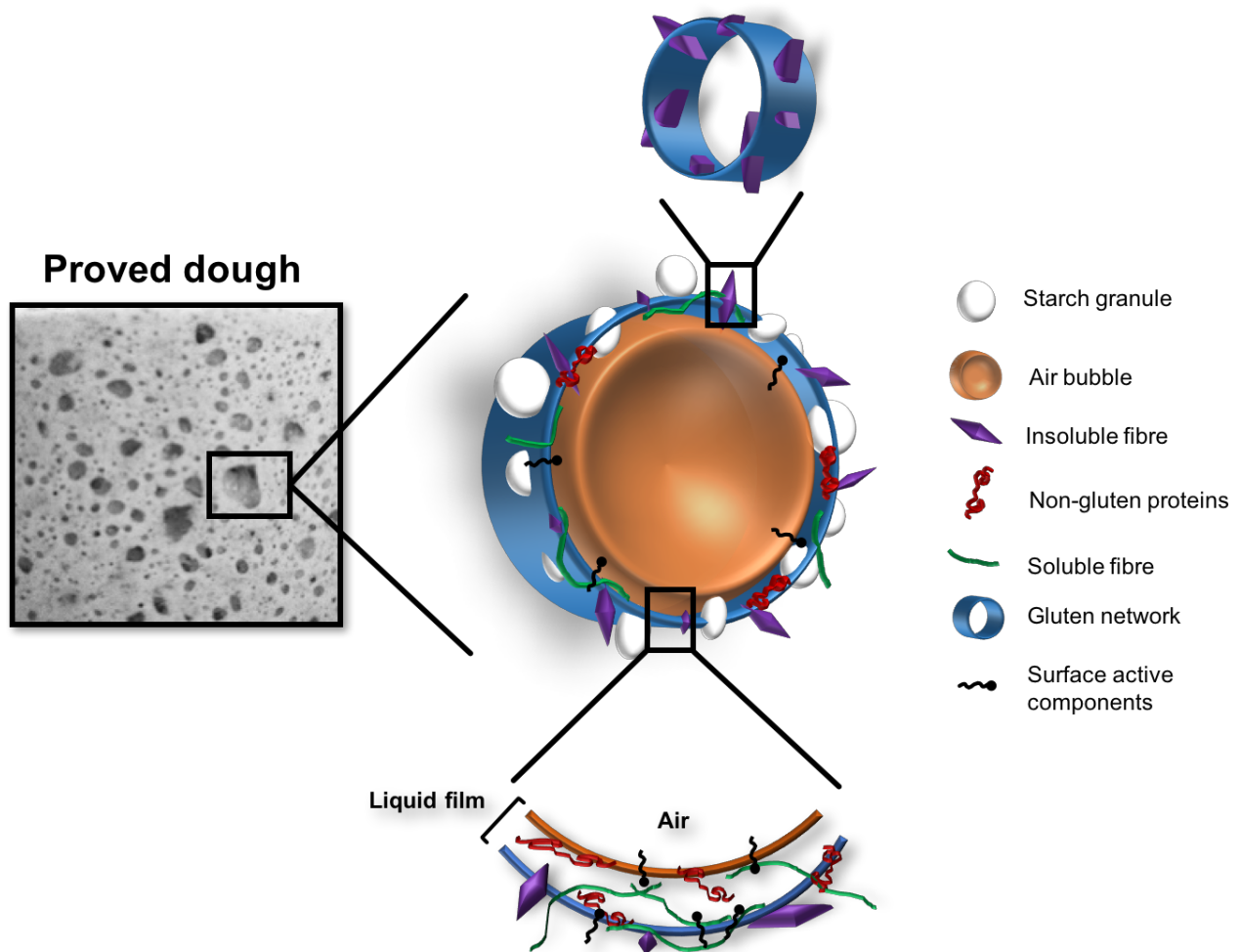


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